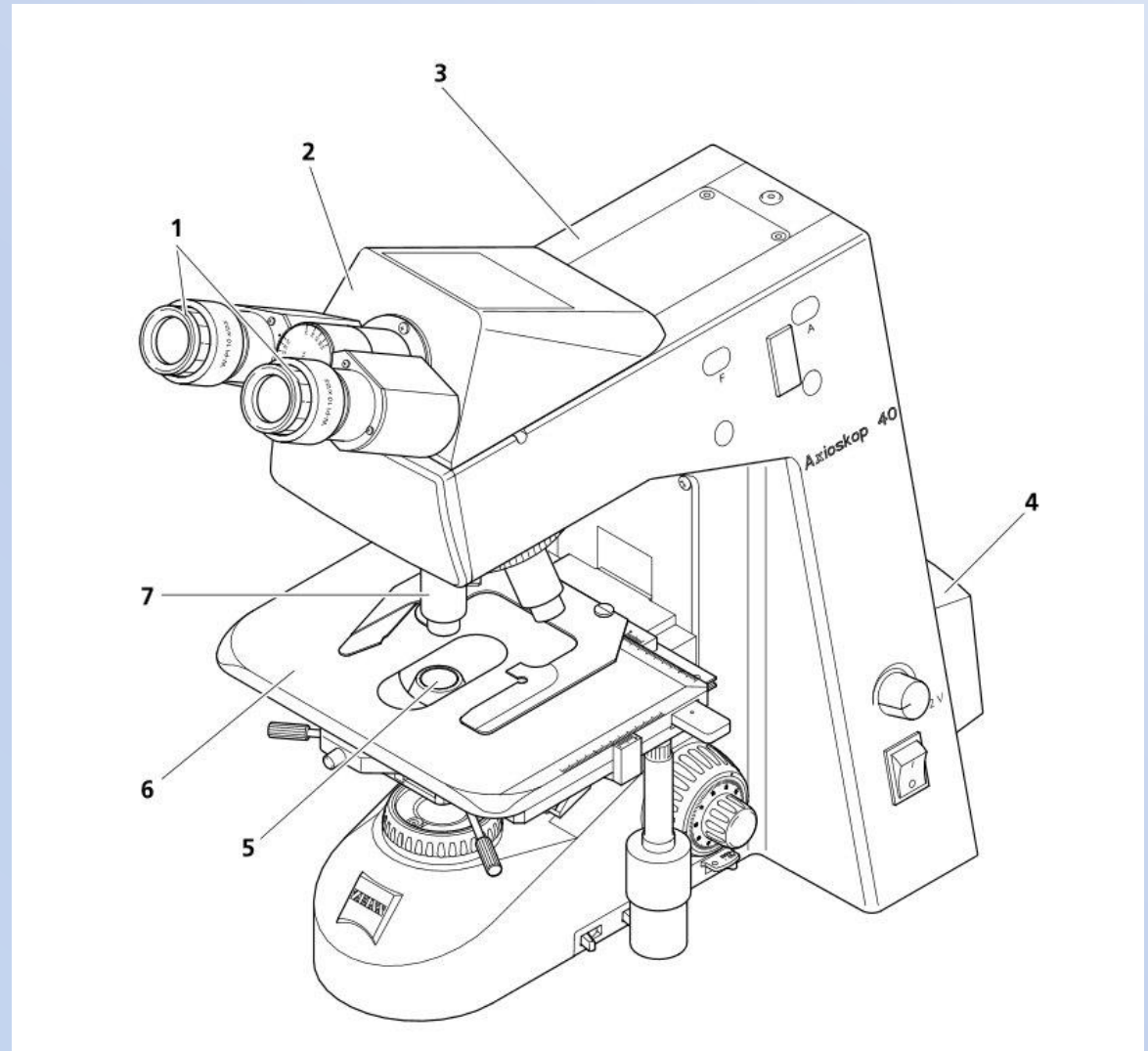


Development of a Remotely-Controlled Autofocus System For a Microscope

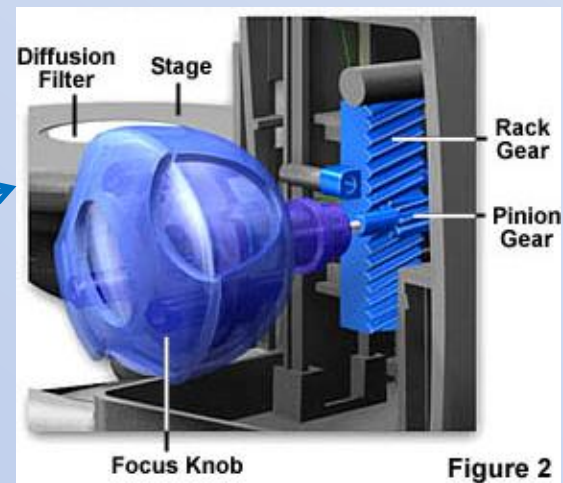
Team members:

1. Athanasopoulos Athanasios
2. Mpantes Fotis
3. Saloufas Michalis
4. Varvagiannis Efstratios



Overview

Automating the vertical motion of microscope's stage

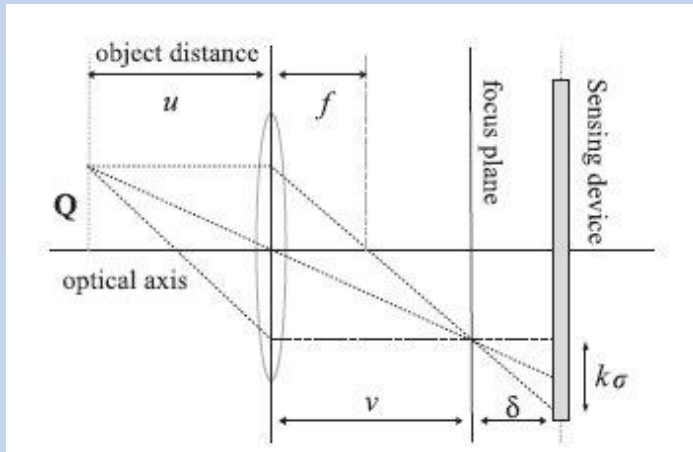


... in our case

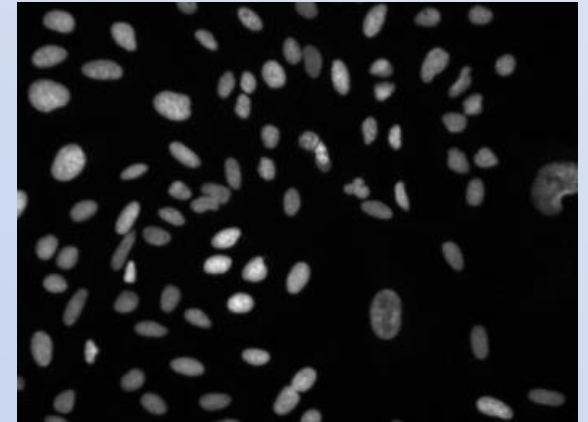


Motivation

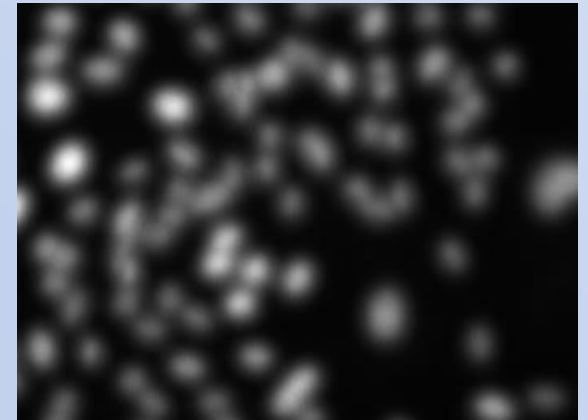
A common issue in microscopy is defocus, especially when imaging thick samples



In focus



Out focus



Auto-focus procedure is a major requirement in automated microscopy

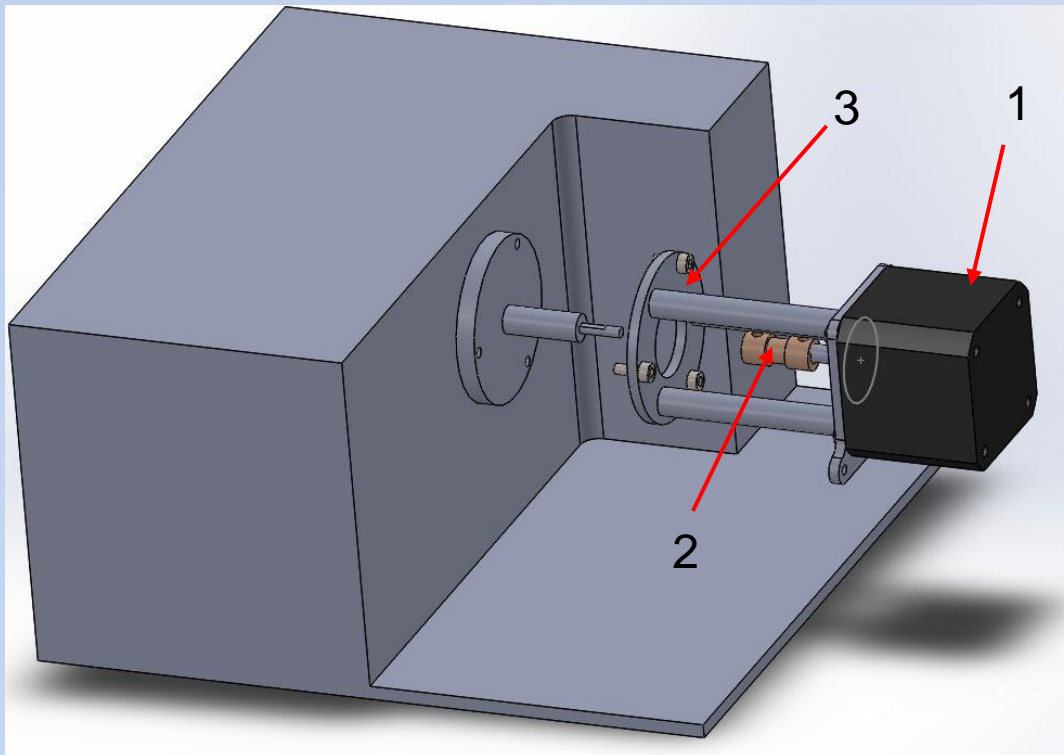
Motivation

- Low-cost implementation of an auto focus procedure
- Wireless transmission of camera data
- Remote control using modern microprocessors

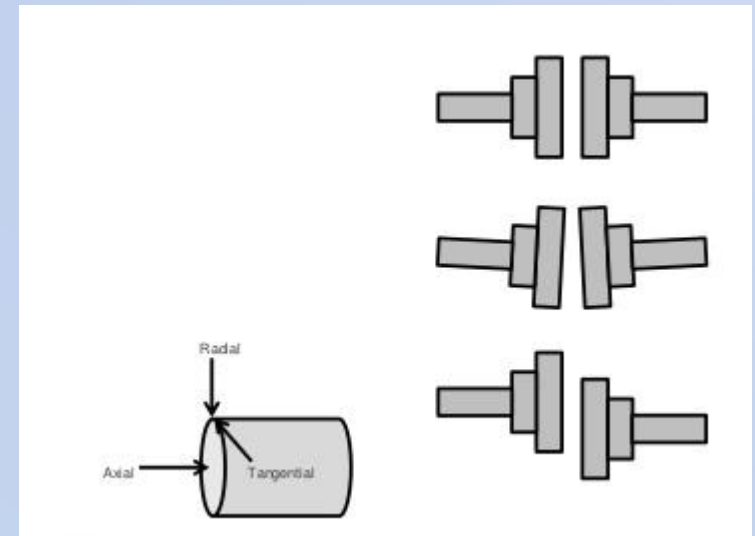
Project Outline

- Mechanical System
 1. Z-axis motion system
 2. Board prototype
 3. Pi camera basement
- Software control
 1. Arduino program
 2. Serial communication
- Remote control via web
 1. Web interface
 2. Image wireless transmission

Mechanical Design

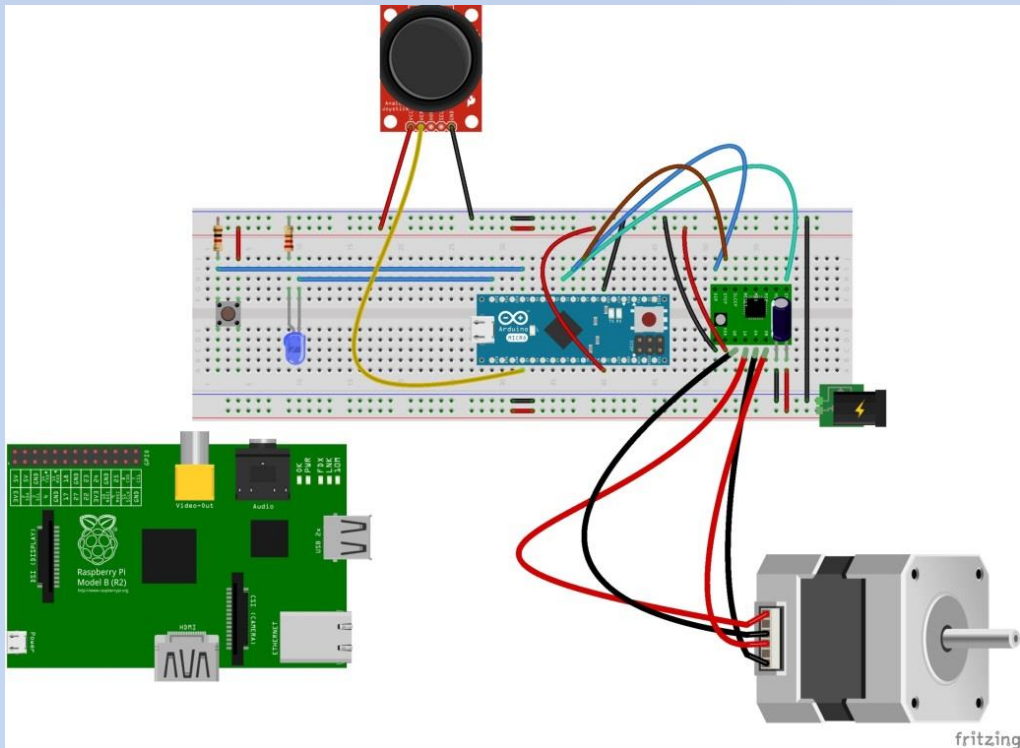


1. **Stepper Motor**
2. **Motion coupler**
 - Attach motor to the microscope
3. **Motor Housing**
 - Ensuring axis alignment

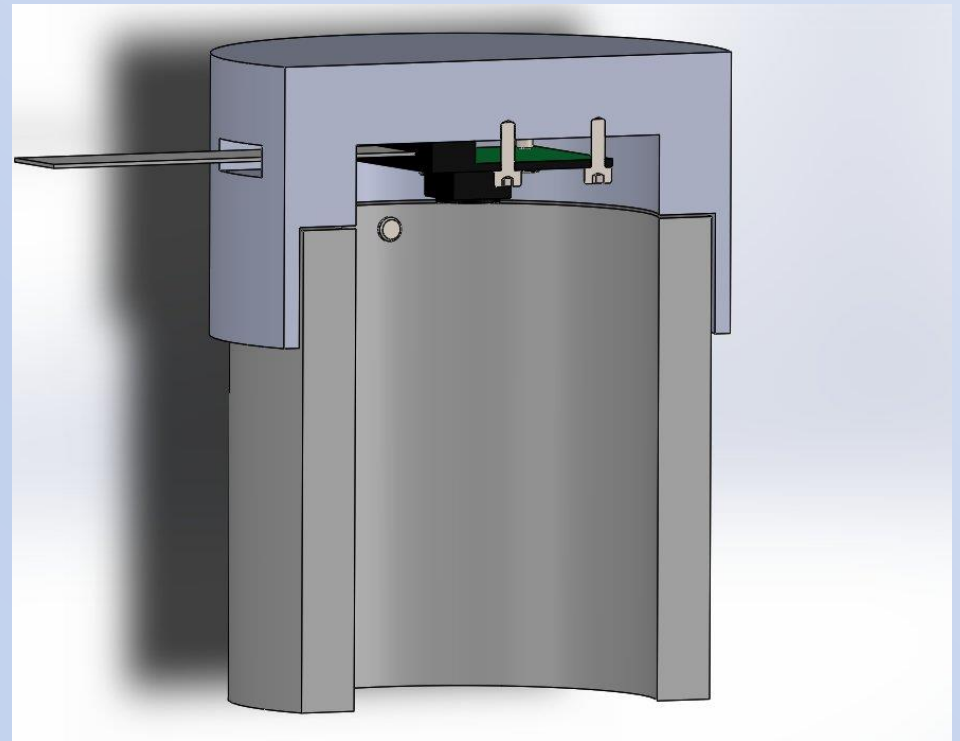


Mechanical Design

- Electronic Circuit



- Camera basement



Software Part

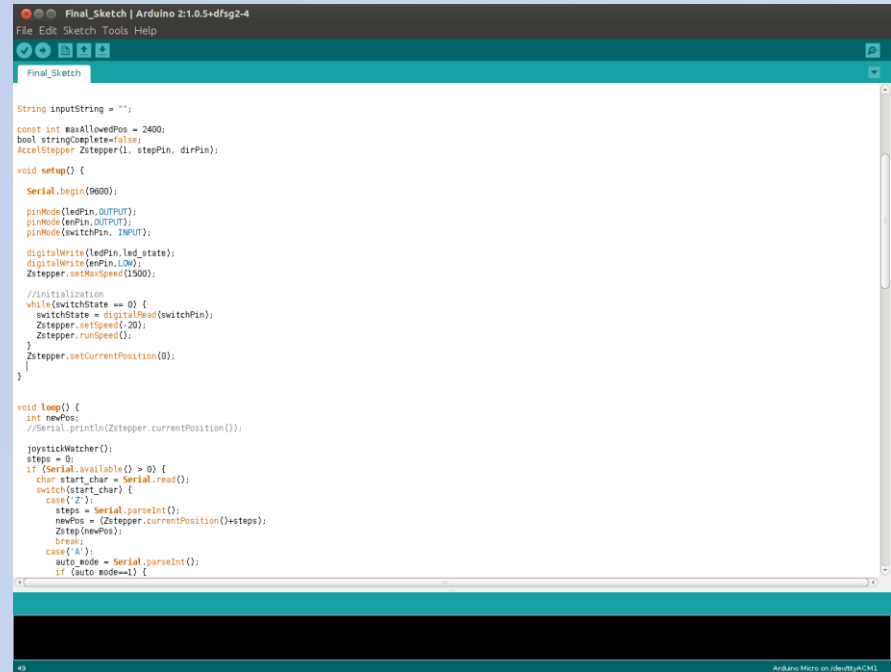
Arduino program

Controls stepper

Communicates with Raspberry
Pi via serial communication
(USB)

Manually with
the usage of a
joystick

From Raspberry Pi



```
String inputString = "";
const int maxAllowedPos = 2400;
bool stringComplete=false;
AccelStepper Zstepper(1, stepPin, dirPin);

void setup() {
  Serial.begin(9600);
  pinMode(ledPin, OUTPUT);
  pinMode(enPin, OUTPUT);
  pinMode(switchPin, INPUT);
  digitalWrite(ledPin, LOW);
  digitalWrite(enPin, LOW);
  Zstepper.setMaxSpeed(1500);

  //initialization
  while (switchState == 0) {
    switchState = digitalRead(switchPin);
    Zstepper.setSpeed(-200);
    Zstepper.runSpeed();
  }
  Zstepper.setCurrentPosition(0);
}

void loop() {
  int newPos;
  //Serial.println(Zstepper.currentPosition());

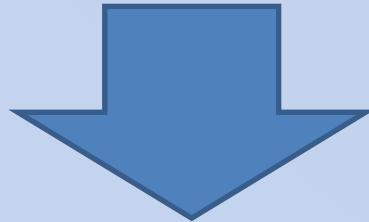
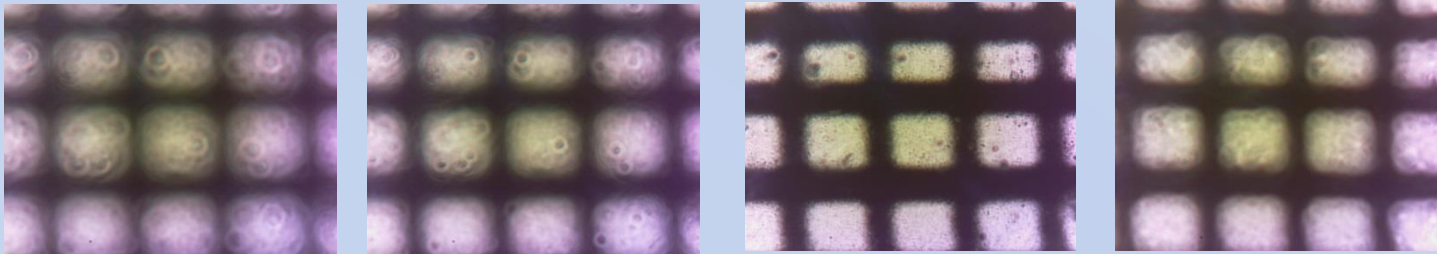
  joystickWatcher();
  steps = 0;
  if (Serial.available() > 0) {
    char start_char = Serial.read();
    switch (start_char) {
      case '2':
        steps = Serial.parseInt();
        newPos = (Zstepper.currentPosition()+steps);
        Zstep(newPos);
        break;
      case '1':
        auto mode = Serial.parseInt();
        if (auto mode==1) {
```


Software Part

On Raspberry Pi (Raspbian- Linux):

- Python scripts for serial communication with Arduino
- Image acquisition from Pi Camera
- Image processing algorithm to detect if an image is blur using

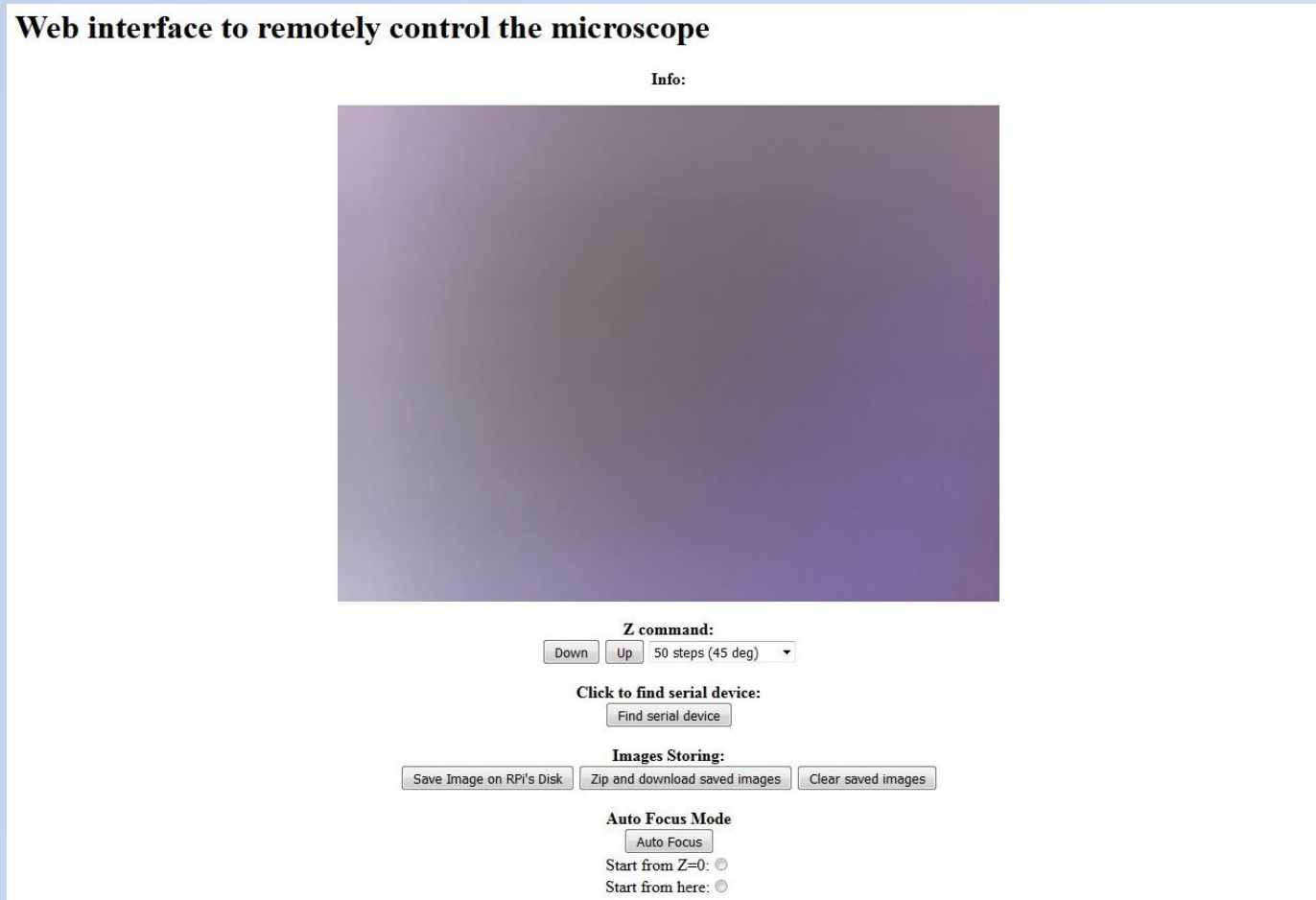
OpenCV library



A simple auto focus procedure

Remote Control

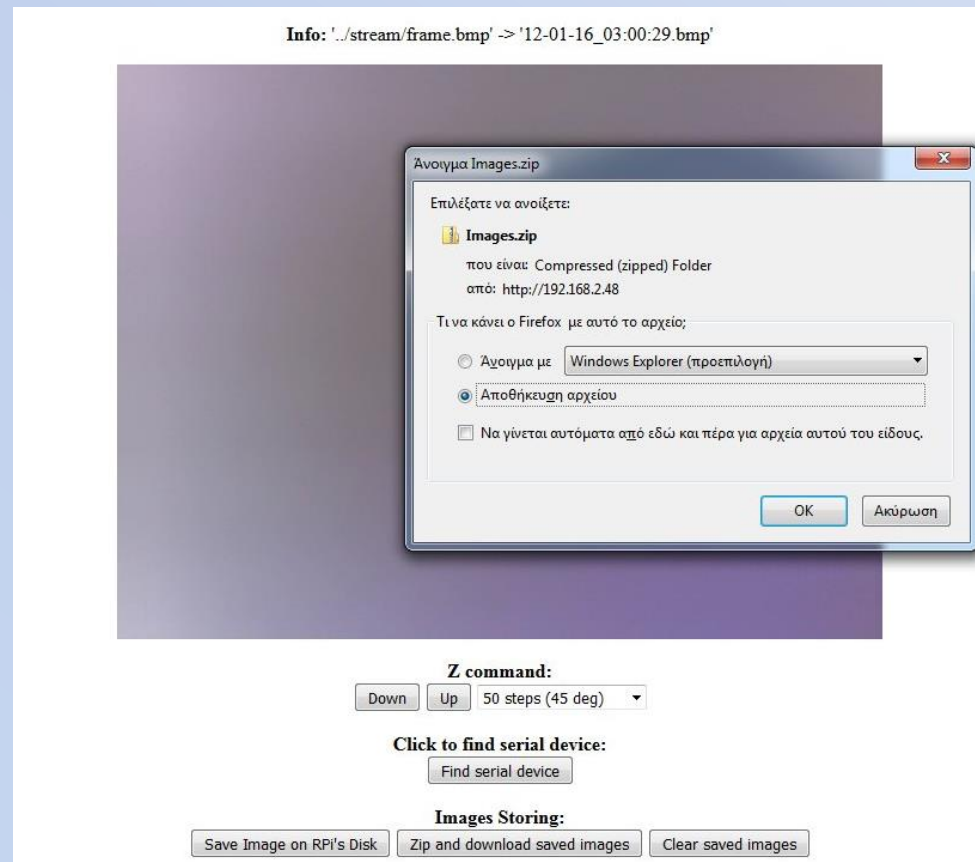
All the scripts written for serial communication with Arduino are accessible via an HTML/PHP page:



This page is available to any device connected to the lab's network by running the Apache server on Raspberry Pi.

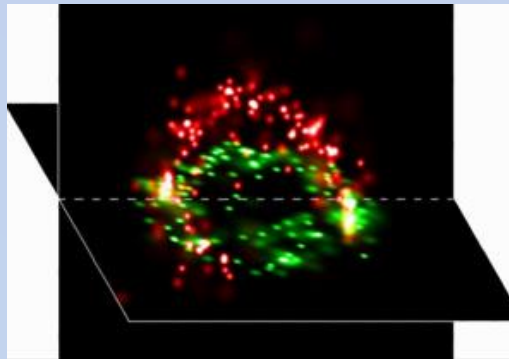
Remote Control

- Live video streaming at 10 fps is available as a sequence of jpeg images using a software called mjpeg-streamer
- Moreover the user can save selected frames and download them in uncompressed format (bmp)



Future Work

- More efficient implementations of this project
- More sophisticated and robust auto focus algorithms
- Full automation of the microscope
 - 3-D microscopy
 - Live microscopy



Summary

Autofocus

Raspberry Pi

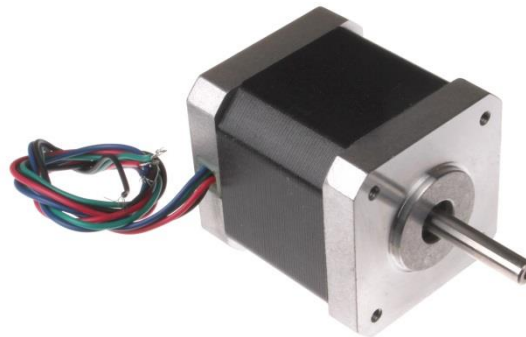
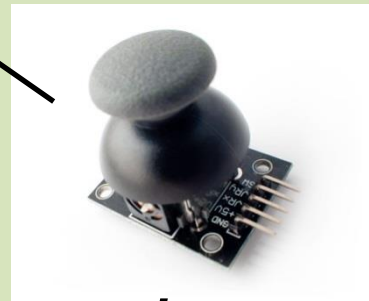
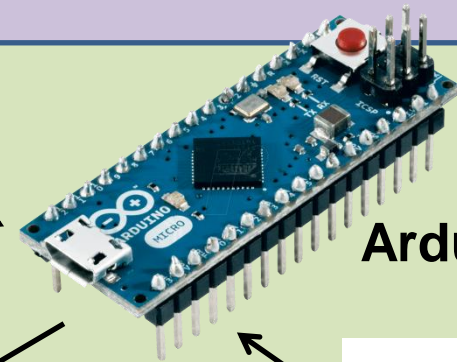
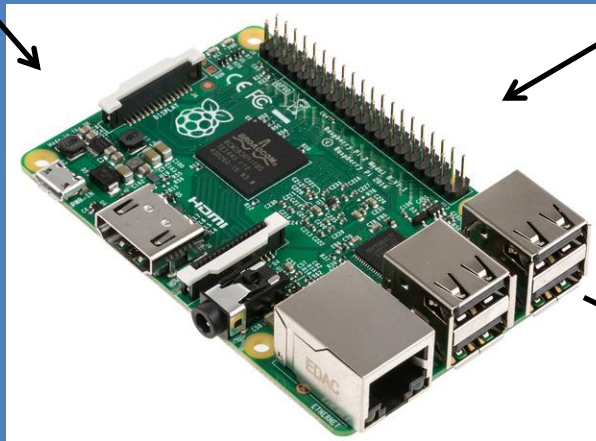
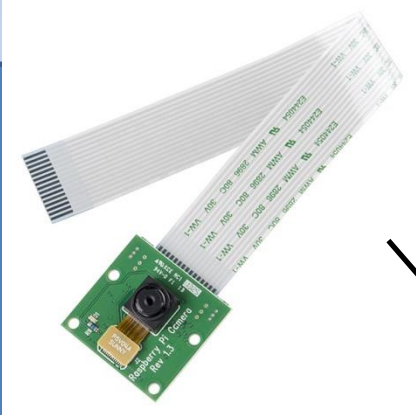
Remote control



Remote user

Arduino mcu

Manual control



Acknowledgments

We would like to thank:

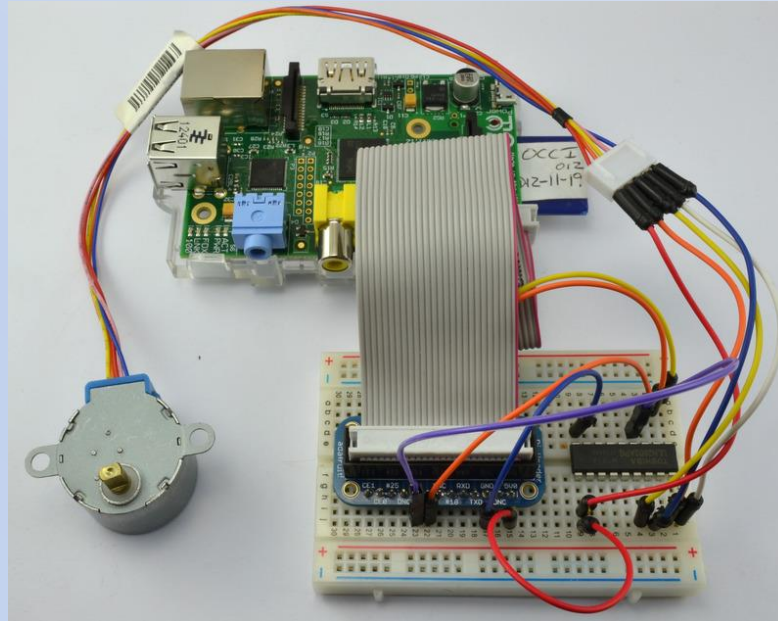
- Mr. Tzeranis Dimitrios for the management of the whole project and his continuous support
- Mr. Polesiounk Alexandros for the construction of the mechanical parts and useful technical advises
- SteelMax for the construction of two flanges free of charge

Thank you!

Questions



Why use arduino?



- In order to control Raspberry's GPIO pins superuser privileges are needed
- ***This is an important security flaw for the raspberry pi system***
- Moreover it is more useful to have a separate controller for the hardware!

Image processing algorithm

- A method consisting in the variance of Laplace operator
- **ADVANTAGE:** Can inform us about the blurriness of an image with a single value
- **DISADVANTAGE:** Is performed on the whole image (convolution with a Laplace kernel)



$\nabla^2 I$
→

Source: Analysis of focus measure operators for shape-from-focus Said Pertuz e.a.

Auto focus techniques

- Digital Autofocus Methods for Automated Microscopy By FEIMO SHEN, LOUIS HODGSON, and KLAUS HAHN
- A Comparison of Different Focus Functions for Use in Autofocus Algorithms Frans C.A. Groen, Ian T. Young, and Guido Ligthart
- Comparison of Autofocus Methods for Automated Microscopy' Lawrence Firestone, Kitty Cook, Kevin Culp, Neil Talsania, and Kendall Preston, Jr
- Blur Detection for Digital Images Using Wavelet Transform Hanghang Tong

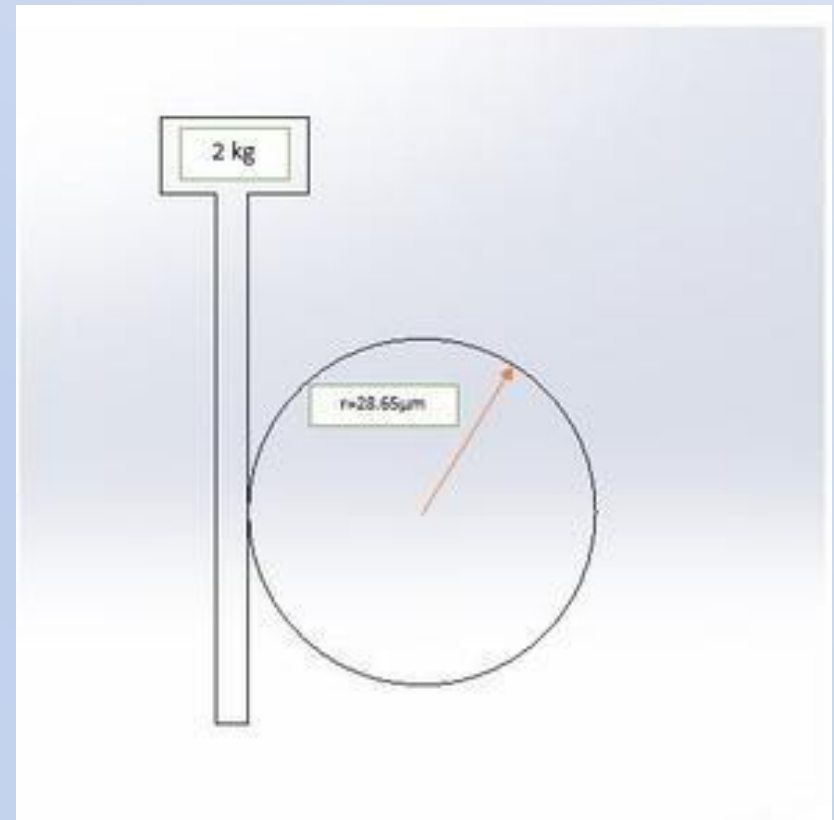
Transmission System Model

1 revolution \rightarrow 180 μm vertical

Torque Required: $4.5 \times 10^{-3} \text{ Nm}$

Calculations speed: 20 rpm

Calculated acceleration: 2,09 m/sec^2
Assuming 1/3-1/3-1/3 accelerating profile.



Stepper Motor

Low speed



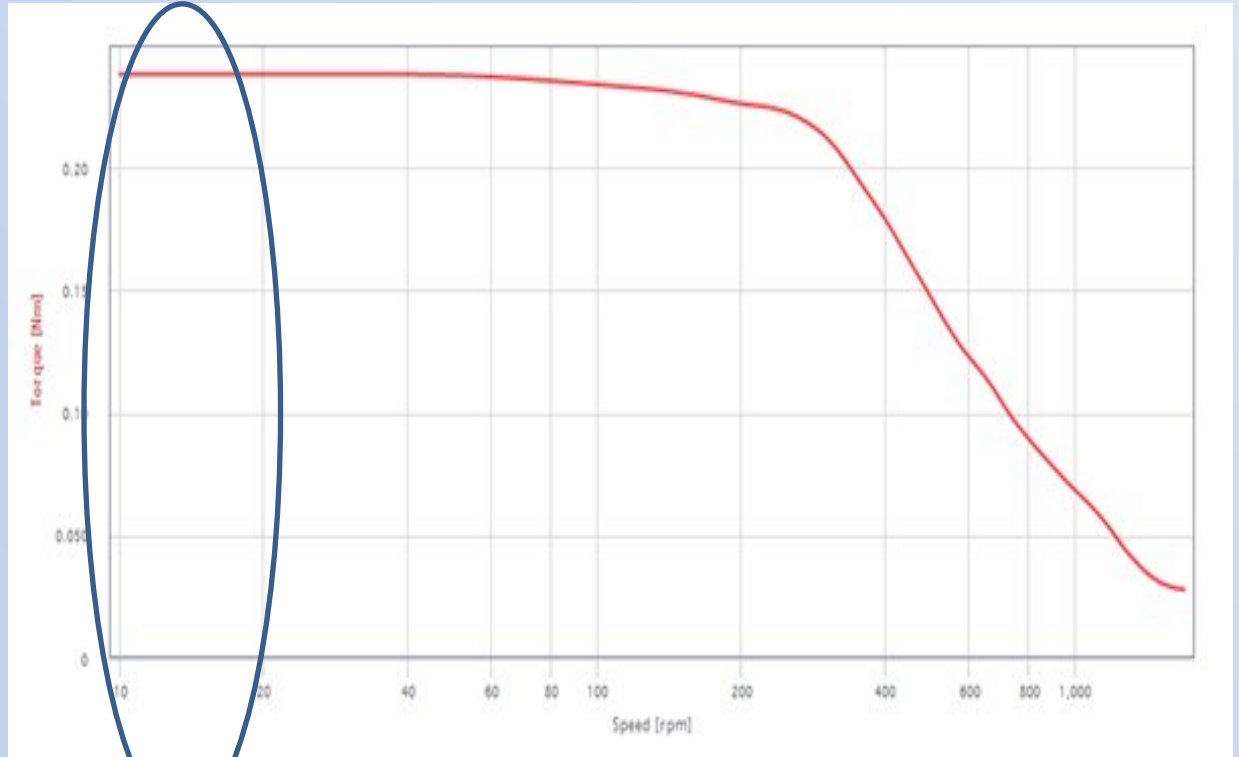
Locked step mode



Deceleration between
each step



Motor not losing steps



Working range